

## Modified Provasoli (MP)

Prepare the following primary stocks solutions first.

Primary Stocks	per litre
(1) $\text{Na}_2 \beta\text{-glycerol PO}_4 \cdot 5\text{H}_2\text{O}$ *	50 g
(2) $\text{NaNO}_3$	35 g
(3) Iron-EDTA:	
$\text{Fe}(\text{NH}_4)_2(\text{SO}_4)_2 \cdot 6\text{H}_2\text{O}$	0.7 g
$\text{Na}_2 \text{EDTA}$	0.6 g
(4) Vitamin B <sub>12</sub>	0.025 g
(5) Thiamine	0.5 g
(6) Biotin	0.05 g
(7) PII trace metals:	
$\text{Na}_2 \text{EDTA}$	1.0 g
$\text{H}_3\text{BO}_3$	1.12 g
$\text{MnSO}_4 \cdot 4\text{H}_2\text{O}$	0.12 g
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	0.022 g
$\text{CoSO}_4 \cdot 7\text{H}_2\text{O}$	0.005 g

To prepare the final stock solution, Using the primary stocks as shown below, make up to 1 litre of deionised water.

Stock	per litre
(1) $\text{Na}_2 \beta\text{-glycerol PO}_4 \cdot 5\text{H}_2\text{O}$ *	8.0 ml
(2) $\text{NaNO}_3$	110 ml
(3) Iron-EDTA	100 ml
(4) Vitamin B <sub>12</sub>	3.5 ml
(5) Thiamine	8.0 ml
(6) Biotin	8.0 ml
(7) PII trace metals	200 ml

Dispense the final stock solution into 10ml aliquots. Autoclave at 15 psi for 15 minutes.

Finally, to use add 20 ml per litre to sterile 30 ppt filtered seawater (950mls filtered seawater: 30mls deionised water)

For ½ strength Modified Provasoli add 10 ml sterile stock per litre of sterile 30 ppt filtered seawater (950mls filtered seawater: 40mls deionised water)

For agar add 15g per litre Bacteriological Agar.

\*  $\beta$ -Glycerol phosphate disodium salt pentahydrate

### Reference

West JA & McBride DL (1999) Long term and diurnal carpospore discharge patterns in the Ceramiaceae, Rhodomelaceae and Delesseriaceae (Rhodophyta). *Hydrobiologia*. **398-399**, 101-114.

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